



The potential of Almond Seed Extract as a substitute for Cow's Milk in the production of Functional Yogurt

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Abstract

Yogurt is a fermented milk product that can be supplemented or substituted with plant extracts, one of which is almond seed extract. This study aims to evaluate the potential of almond seed extract (*Prunus dulcis*) as a substitute for cow's milk in the production of functional yogurt. The almond seed extract, which is rich in bioactive compounds and antioxidants, is used in yogurt formulations (almond extract: cow's milk) with ratios of 25:75, 50:50, and 75:25, with the measured parameters being pH, color, and water holding capacity (WHC). The obtained data were analyzed using analysis of variance and Duncan's multiple range test. The research results indicate that increasing the proportion of almond extract significantly lowers the pH of yogurt, with the lowest value at a 75:25 ratio (pH 4.47). Color analysis shows a decrease in brightness (L^*) and an increase in greenness (a^*) and yellowness (b^*) at higher almond concentrations. The highest L^* (84.86), a^* (-3.12), and b^* (9.46) values were obtained at the 75:25 ratio. Water holding capacity (WHC) decreased with the increase of almond extract, with the highest WHC at the 25:75 ratio (55.1%). with the substitution of almond seed extract improving the functional properties of yogurt, and the 25:75 substitution being an optimal combination with a stable texture and yogurt product.

Keywords: Almond extract, colour, WHC, yogurt

Introduction

Yoghurt is one of the fermented dairy products. Milk is fermented using lactic acid bacteria such as *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. Yoghurt is known as a functional product due to its probiotic content and high nutritional value, especially as a source of protein, calcium, and B-complex vitamins (Tamime & Robinson, 2007; Granato *et al.*, 2010) [4, 11]. Along with the public's awareness of consuming functional foods, diversification of dairy products is being carried out by substituting with plant extracts (Wulandari *et al.*, 2022) [13].

One of the plant-based ingredients or extracts that has potential for use is almond seed extract (*Prunus dulcis*), which is obtained through the extraction process from whole almond seeds. Almond seed extract is known to contain bioactive compounds such as unsaturated fatty acids, phenols, flavonoids, and vitamin E, which act as antioxidants and have beneficial health effects (Mandalari *et al.*, 2010; Barreca *et al.*, 2016) [2, 8]. The results of several studies indicate that almond extract has potential as a functional ingredient in food formulations due to its antimicrobial and anti-inflammatory properties, as well as its ability to support metabolic health (Hartati *et al.*, 2019b) [5, 6] Chen *et al.*, 2006; Ros *et al.*, 2010) [3, 10].

The use of almond seed extract as a substitute ingredient in yogurt also reduces lactose levels and contributes to an increase in bioactive compound content in the final product. However, the differences in chemical and functional characteristics between cow's milk and almond seed extract. the differences lie in the composition of protein and fat, which can affect the texture, pH, microbial starter viability, and organoleptic characteristics of yogurt (McClements, 2020; Jeske *et al.*, 2018) [9]. Therefore, this study aims to evaluate the potential of almond seed extract as a substitute for cow's milk in the production of functional yogurt. The results of this study are expected to serve as a foundation for

the development of high-nutritional-value plant-based fermented products, safe for consumption by consumers with restrictions against animal milk, and in line with the trends of healthy and sustainable food.

Preparation of Yogurt Almond

Almond extract and fresh cow's milk in the ratios of 25:75, 50:50, and 75:25 are pasteurized at a temperature of 75°C for 15 minutes. After pasteurization, the mixture of milk and almond extract is removed and cooled by placing the pot into a basin or container filled with water while stirring gently until the temperature reaches 43°C. Once the milk temperature reaches 43°C, the yogurt culture is poured into each pot containing the milk. The milk and yogurt culture are stirred until they are evenly mixed. The milk mixed with the yogurt culture (*Lactobacillus bulgaricus*, *Streptococcus thermophilus* and *Lactobacillus acidophilus*) is transferred into a clean container. The container is tightly closed to prevent air from entering, allowing the incubation process to proceed perfectly. The mixture of milk and yogurt culture is left to sit so that the bacteria can grow and develop. After being left to sit or incubated for 12-15 hours, the resulting yogurt is ready to be analyzed.

pH

The measurement is conducted by immersing the pH meter electrode into 10 ml of the sample (AOAC, 1995)

Colour

Color measurement (L , a , and b) was conducted using a colorimeter with the Engelen (2018) method by determining the red, blue, yellow, and green colors of the light. The light is absorbed by the sample. The sample was prepared in a 250 ml glass jar.

The L* value as a parameter of achromatic color brightness ranges from 0-100. The a* value as a parameter of chromatic color is a* = 0-100 for red and a- = 0-(-80) for green. The b* value as a parameter of chromatic color is b* = 0-70 for yellow and b- = 0-(-70) for blue.

Water holding capacity

The water holding capacity (WHC) test was conducted using centrifugation based on the method from Clunies *et al.* (1986). A sample of 1 g was added to 10 mL of distilled water. The suspension was then stirred for 5 minutes and placed in a centrifuge tube. Next, the sample is centrifuged for 30 minutes at a speed of 3500 rpm. Supernatant was separated and the sample precipitate was weighed.

Result and Discussion

pH value

The results of the study on the substitution of cow's milk with almond seed extract are shown in Table 1. With the addition of almond extract in yogurt, the pH of the resulting yogurt significantly decreased.

Table 1: pH Value of yogurt

Treatment	pH
25: 75	4,84 ^a
50: 50	4,57 ^b
75: 25	4,47 ^c

The difference in pH values can be attributed to the fermentation activity of lactic acid bacteria (LAB), which serve as the starter cultures for the yogurt, namely *Lactobacillus bulgaricus*, *Streptococcus thermophilus* and *Lactobacillus acidophilus*. These LAB play a role in converting lactose into lactic acid. The substrate composition—specifically the levels of protein, fat, and carbohydrates—greatly influences microbial growth, acid production, and also affects the fermentation rate and final outcome, including the pH (Wulandari *et al.*, 2024) ^[12].

In the 50:50% ratio of cow's milk and almond extract, it is suspected that there is an optimal interaction between almond components (such as phytonutrients, fiber, and non-lactose sugars) and milk components, which supports more active growth of lactic acid bacteria (LAB), resulting in a lower pH. This is supported by a study conducted by (Hartati *et al.*, 2019a) ^[5, 9], which found that the addition of plant extracts can enhance the availability of substrates, thereby accelerating the decrease in pH during the fermentation process.

Color Characteristics

The results of the study on the color characteristics of yogurt with various ratios of almond extract and cow's milk (25:75, 50:50, and 75:25), including lightness (L*), red-green value (a*), and yellow-blue value (b*), are presented in Table 2.

Table 2 Color characteristic of Yogurt

Treatment	L	a*	b*
25: 75	81.11 ^b	-2.4 ^a	8.38 ^b
50: 50	81.61 ^b	-2.78 ^a	8.59 ^b
75 : 25	84.86 ^a	-3.12 ^b	9.46 ^a

From the obtained data, the L value ranges from 84.86 to 81.11. In the 75:25 ratio, it shows the highest brightness (84.86a) and is significantly different (p < 0.05) from the other treatments. The L value decreases with the addition of almond extract, and the color of the yogurt becomes darker. This is suspected to be caused by the presence of natural pigments in almond extract, namely polyphenols and flavonoids, which affect the color of the product. From Table 2, the a* value shows negative numbers for all treatments, indicating that the yogurt has a tendency towards a greenish color. There is no significant difference between the 25:75 (-2.4a) and 50:50 (-2.78a) treatments, but the 75:25 formulation shows a lower a* value (-3.12b), which means a more intense greenish color and a significant difference (p < 0.05). This research aligns with the study conducted by Chen *et al.* in 2006 ^[3], which found that almond extract can enhance the greenish hue due to the phytochemical compounds it contains.

Meanwhile, the b* value, which indicates the intensity of the yellow color, is highest in the 75:25 formulation (9.46) and significantly different from the 25:75 and 50:50 treatments. The increase in b* value indicates that the higher the concentration of almond extract, the more intense the yellow color in the yogurt, likely due to the yellowish-brown pigments from the almond skin and oil. This finding is consistent with the research by Mandalari *et al.*, (2010) ^[8] which stated that bioactive compounds from almonds contribute color to food products.

Overall, the increase in the proportion of almond extract in the yogurt formulation significantly affects the color attributes, namely reducing brightness and increasing the intensity of green and yellow colors. These changes are important to consider in the development of plant-based yogurt to maintain sensory appeal while also providing functional benefits.

Water Holding Capacity

Water Holding Capacity (WHC) describes the ability of yogurt to retain water within its gel matrix without releasing free liquid (whey). Water holding capacity for yogurt with cow's milk substituted with almond extract is presented in Table 3.

Table 3 Water holding capacity of Yogurt

Treatment	WHC (%)
25: 75	55.1 ^a
50: 50	52.3 ^b
75: 25	52.9 ^c

Based on Table 3, it is shown that the substitution between almond extract and cow's milk has a significant effect on the water holding capacity (WHC) of yogurt. The WHC values range from 52.9 to 55.1, and the WHC values differ significantly between treatments.

The WHC value indicates the ability to retain water in the gel matrix, which is closely related to the strength of the protein structure formed during the fermentation process (Arab *et al.*, 2023) ^[1]. The higher casein content in cow's milk results in a denser and stronger gel structure at the 25:75 ratio, allowing it to retain water more effectively. The decrease in WHC at the 50:50 and 75:25 ratios indicates that the increased proportion of almond extract reduces the gel's ability to bind water. This may be due to the lower quantity and quality of protein in almond extract, which does not

support the formation of a strong gel network as well as milk protein. Although the 75:25 treatment has a higher proportion of almond extract compared to the 50:50 treatment, its WHC value is slightly higher but still statistically significantly different, indicating that the interaction between plant and animal proteins can produce different gel structures. Thus, the yogurt formulation with a 25:75 ratio is the most optimal in retaining water, contributing to a thicker texture and better product stability. The decrease in WHC at higher almond ratios could be an important consideration in the development of plant-based yogurt.

Conclusion

The substitution of cow's milk with almond seed extract up to 75% reduced the pH, resulting in decreased brightness, with color characteristics of L* 84.86, a* -3.12, and b* 9.46, and a WHC value of 52.9%.

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